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Ecology Of Larra Bicolor (Hymenoptera: Sphecidae) In The Northern Gulf

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ECOLOGY OF *LARRA BICOLOR* (HYMENOPTERA: SPHECIDAE) IN THE

NORTHERN GULF

By

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ECOLOGY OF *LARRA BICOLOR* (HYMENOPTERA: SPHECIDAE) IN THE NORTHERN GULF

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Mole crickets (*Scapteriscus* spp.) are the most destructive pests in southern turf and pasture grasses. In response to extensive losses from mole crickets, Florida formed a task force to identify natural enemies in the native range of these pests. Two parasitic insects, *Larra bicolor* and *Ormia depleta,* and *Steinernema scapterisci,* an entomopathogenic nematode, were imported and released. Of the two insects, only *Larra bicolor* has spread to other states infested with mole crickets in the southeast. The present study documents the seasonal biology of *Larra bicolor* in the northern Gulf region, ornamental plants that can be used as nectar sources, and the impact of these nectar sources on longevity of the wasp and parasitism of mole crickets. Results of field and laboratory experiments showed that the ornamenal *Pentas lanceolata* attracted wasps in the field and provided comparable or better longevity than *Spermacoce verticillata* which was the only known nectar source.

Key words: *Larra bicolor*, nectar sources, Mole cricket Bio control, *Spermacoce verticillata*, *Scapteriscus* control.

DEDICATION

I would like to dedicate this document to my parents, M. C. Abraham and Elizabeth Abraham, brother Tomci and fiancée, Tina.

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CHAPTER I

GENERAL INTRODUCTION

Introduction to Turfgrass

Of the 7500 species of plants in family Poaceae (= Graminae), only about 40 species are used for the various functions of turf (Tashiro 1987). The commonly used term, 'turfgrass' refers to the plant of grass, while 'turf' represents a community of turfgrasses including the medium of its growth (Turgeon 2005). According to the climatic differences, turfgrasses are mainly divided into warm season grasses and cool season grasses. They differ in their requirement of climatic condition, photosynthetic efficiency and other growth related processes. Warm season grasses (C4 plants) require 27–35°C (80–95°F) for optimum growth, whereas cool season grasses (C3 plants) require 16–24°C (60–75°F) for optimum top growth and 4–16°C (40–60°F) for root growth (Turgeon 2005). Although C4 plants are considered more photosynthetically efficient, C3 plants show limited or no winter dormancy (Turgeon 2005). The difference in the growth pattern of these two grass types is shown in Figure 1.1 and 1.2.

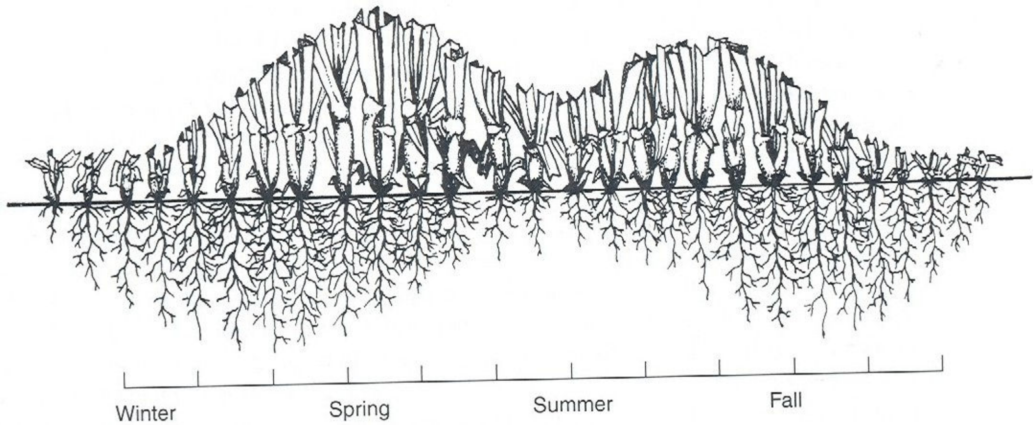

Figure 1.1

Growth Pattern of Cool Season Turf (Turgeon 2005)

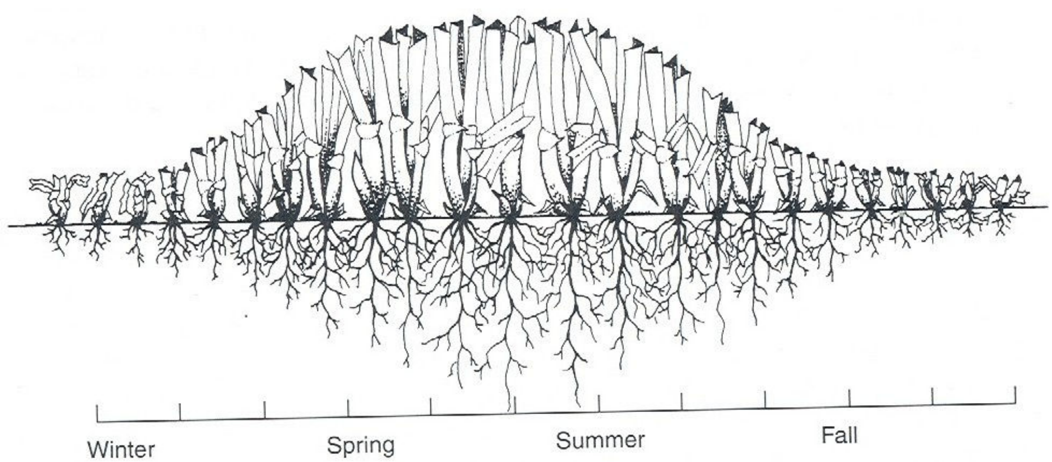

Figure 1.2

Growth Pattern of Warm Season Turf (Turgeon 2005)

According to climate and precipitation, the continental U. S. is divided into six turfgrass zones (Potter 1998). Mississippi is in zone 3 and comes under the warm humid zone. Even though the entire state is classified under zone 3, the climatic conditions do vary along the length and breadth of the state. The area along the Gulf Coast experiences a subtropical climate which is warm and humid, but places north of the coast experience cooler winters and warmer summers. The tropical climate in the coastal region makes it best for the growth of warm season grasses. Bahiagrass (*Paspalum notatum)* , bermudagrass (*Cynodon spp.*), zoysiagrass (*Zoysia* spp.), St. Augustinegrass (*Stenotaphrum secundatum*), and centipedegrass (*Eremochloa spp.*) are the main grasses grown in this region. Conditions in the northern part of Mississippi are more favorable for zoysiagrass, whereas conditions in the southern part of the state are more suited to growth of bahiagrass and St. Augustinegrass (Tashiro 1987). Centipedegrass has distribution all over the state mostly in home lawns while bermudagrass is used in golf courses and other areas.

Unlike row crops and other plants of commercial importance, turfgrass and its importance are often overlooked. Yields and losses in turfgrass are not as we usually see in traditional crops or cropping systems. Turfgrass has more of an aesthetic value and its benefits are not often tangible. For example, here are a few qualities of turf which make them important (Morris 2003):

- Moderately sized lawns of eight homes are capable of cooling air equivalent to 70 tons of air conditioning.

- A 232.3 m^2 (2500 $foot^2$) lawn absorbs carbon dioxide and produces enough oxygen to support a four member family.
- A healthy lawn reduces surface water runoff up to 80 times.
- A beautiful lawn increases home property values by 15%.

Apart from these statistics, turfgrass beautifies millions of home lawns, is a safe playing surface on over 700,000 athletic fields, and provides a source of outdoor recreation for over 26 million golfers on about 17,000 golf courses. With tens of thousands of sod producers, lawn care operators and landscapers, there are at least 20 million ha (50 million acres) of managed turf, with an estimated $45 billion industry in all forms in the U. S. (Beard and Green 1994). In Mississippi, there are more than 1 million ha (2.5 million acres) of maintained turf, with at least 121,405 ha (300,000 acres) of lawn surrounding 740,000 homes, 160 golf courses and 2,000 athletic fields (Anonymous 2005). For this reason, turfgrass is an economically-important industry.

Turfgrass Pest Management

According to the functions turf performs, the tolerance level to pests and diseases vary. The tolerance to pests on low budget golf courses (i.e., municipal or public golf courses) would be more than that of private or semi-private golf courses. Similarly, the tolerance level for pests in home lawns differ from that of an athletic field. Private or semi-private golf courses would have little or no tolerance for pests and typically use more chemicals to control pests than public golf courses.

Pests and diseases vary according to the climatic zone in which the turf is grown. For example, mole crickets are commonly agreed upon as the most menacing pest in turfgrass in the southern or southeastern parts of the U.S., but of no concern in the northern U.S. where white grubs are more problematic. Apart from fertilizers, annual expenditures on turfgrass are mostly for plant protection chemicals such as herbicides, fungicides, and insecticides. Despite an emphasis on integrated pest management, turf managers may be more likely to choose a chemical control than risk losing a fairway or putting green. As evidence of this, 125,000 ha of high maintenance areas of golf courses like tees, greens, and fairways were sprayed with insecticides in 1996 (Racke 2000). By comparison, 31% of pesticides used by lawn care companies are insecticides versus only 19% on golf courses (GCSAA 1998, Racke 2000). The targets of these insecticides, particularly in the warm humid region are pest mole crickets (*Scapteriscus* spp.), chinch bugs (*Blissus* spp.), and caterpillar pests like sod webworms, armyworms, cutworms, and fire ants.

Mole Crickets

"Mole crickets are the most destructive insect pests of golf courses, lawns and sod farms in the southeastern United States. These pests account for hundreds of millions of dollars in damage and control costs every year" (Potter 1998). Extensive areas planted in bahiagrass and bermudagrass, accompanied by sandy soils which aid and favor their tunneling behavior (Tashiro 1987), make the Gulf and Atlantic coasts best suited for mole crickets.

Pest Species, Distribution and Origin

Of the 80 species of mole crickets worldwide, ten occur in continental U. S., Hawaii, Puerto Rico, and Virgin Islands namely; *Neocurtilla hexadactyla* (Perty) the northern mole cricket, *Gryllotalpa cultriger* Uhler the western mole cricket, *Gryllotalpa major* Saussure the prairie mole cricket, *Gryllotalpa gryllotalpa* (L.) European mole cricket, *Gryllotalpa orientalis* Burmeister oriental mole cricket, *Scapteriscus abbreviatus* Scudder shortwinged mole cricket, *Scapteriscus borellii* Giglio-Tos (=*S.acletus*) southern mole cricket, *Scapteriscus didactylus* (Latreille) West Indian mole cricket, or changa, *Scapteriscus imitatus* Nickle and Castner imitator mole cricket, and *Scapteriscus vicinus* Scudder tawny mole cricket (Nickle 1992). Among these, only four species are common in the southeastern U. S.; the northern mole cricket, a native species, and the immigrant species; *Scapteriscus vicinus*, *S. borellii*, and *S. abbreviatus*. The northern mole cricket is a four clawed mole cricket unlike the other three which are two clawed (Frank et al. 1994). The northern mole cricket is one of 35 species of four clawed mole crickets occurring in three genera *Neocurtilla, Gryllotalpella, Gryllotalpa* (Nickle and Castner 1984).

Short-winged and southern mole crickets, were introduced into Brunswick, Georgia in 1904, whereas tawny mole cricket was introduced half a decade earlier at the same place (Walker and Nickle 1981). They are believed to have immigrated to the U. S. by hitchhiking in the ballasts of the ships, after which they slowly spread. There are two morphologically recognizable forms of the southern mole cricket, four-dot form and mottled pronotum form, which were introduced separately. The form with a mottled pronotum entered the U.S. through South Carolina in 1916, Texas in 1926, and Florida in 1920 (Nickle and Castner 1984). Since their wings are vestigial, short-winged mole crickets cannot naturally spread far and are mostly problematic in southern Florida. In Mississippi, the southern and tawny mole crickets occur in the southern part of the state typically south of I-20 (Held, personal observation).

Dactyl spacing is a main character which helps distinguish between *S. vicinus* and *S. borellii.* The two tibial dactyls on *S. borellii* are separated by almost the same width as either of the two claws, whereas the tibial claws on *S. vicinus* are separated by a smaller distance giving an apparent V-shape to the tawny mole cricket and a U-shape to the southern mole cricket (Hayslip 1943) (Figure 1.3).

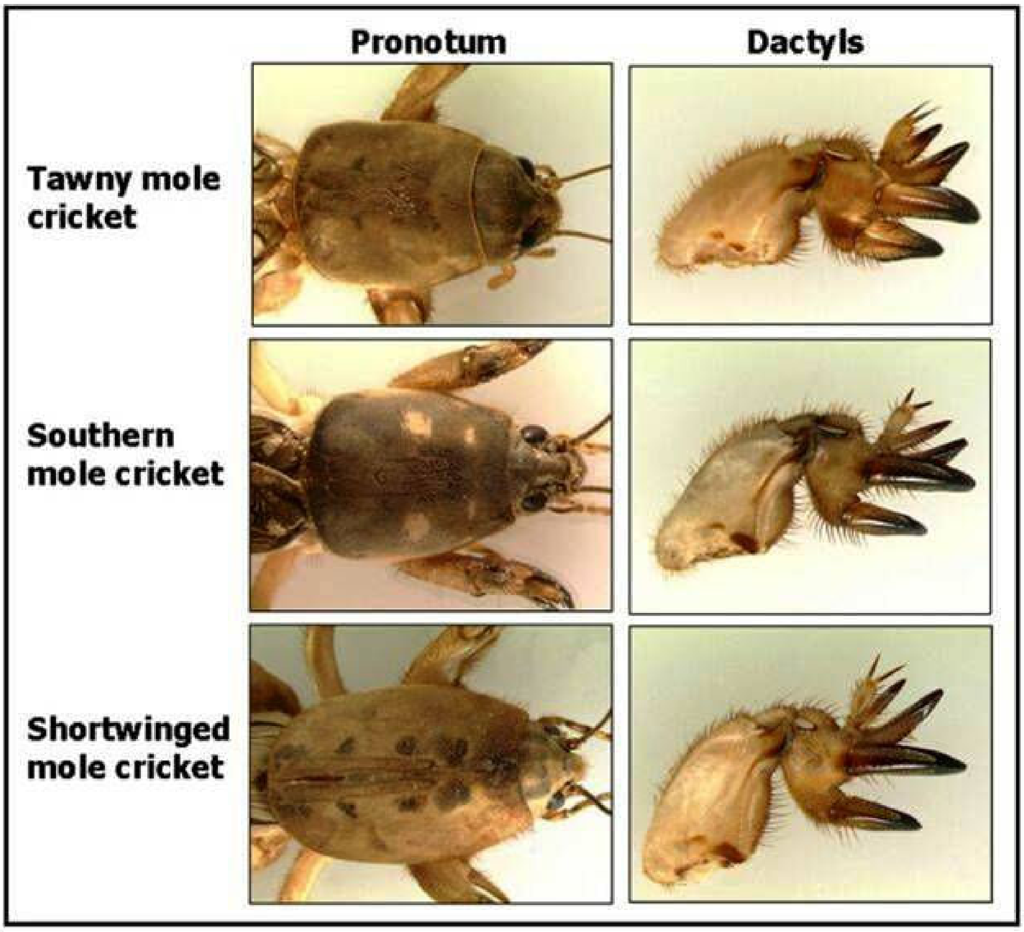

Figure 1.3

Differences in Dactyls Spacing and Pronotum Coloration in *Scapteriscus* Mole Crickets (photo credit, L. Buss, Univ. of FL).

Damage by Mole Crickets

With their strong front legs (fossorial), mole crickets cause three types of cavities in the ground. Tunnels refer to the deeper mines they make in the ground. Galleries are the horizontal mines made just below the soil surface which cause soil to bulge above the surface. Both sexes can make tunnels and galleries, but egg chambers are specialty cavities made by gravid females. They occur at depths varying from 10-30 cms below the soil surface. So, in the soil profile beginning at the surface, we would encounter galleries first, then tunnels, and later egg chambers. Galleries are damaging in golf course putting greens because they deflect the roll of a golf ball. Heavy rains and irrigation can clear these galleries in sandy soils so fresh damage is often apparent.

Apart from the tunneling, herbivory can also cause damage (Frank et al. 1994). Gut examination of the southern mole crickets show that their diet mostly (91%) constitutes of insects, whereas tawny mole crickets have a plant-rich diet (66%) with insects or animals constituting a smaller portion (Taylor 1979). Other *Scapteriscus* mole crickets are similar in feeding habits and mainly fall into these two categories (Castner and Fowler 1984).

Root disruption through tunneling causes drying of the soil and uprooting followed by desiccation of infested grass. Excessive tunneling can weaken or kill a stand of turfgrass (Adjei et al. 2003). Even though species may differ by the characteristic damage they make, all species do cause mechanical damage (Castner and Fowler 1984). Increased damage is accentuated in moist (Hertl and Brandenburg 2002) and sandy soils. As a result, well-watered turfgrass, especially on golf courses, are more vulnerable to mole cricket damage (Hayslip 1943).

Chemical Control

Being subterranean pests, mole crickets are not easy to control. To be effective, chemicals must penetrate the thatch layer and reach the mole crickets in a dose which would be lethal to them. This may require multiple applications of pesticides to keep them under check. Mole crickets can reportedly detect and avoid soils treated with insecticides (Brandenburg 2002) further confounding management. We can observe large amounts of damage during the spring, therefore chemical applications are necessary at that time, however the efficacy of pesticides are impeded by the large nymphs and adults abundantly present at that time. Since the insecticide would be more effective against younger nymphs, they are to be targeted for optimum results. This gives a window from June through July for application of the pesticides, because nymphs are small and easily controlled but damage is not obvious. This period however, is the best chance to control mole crickets (Held and Layton 2006). Baits are insecticides formulated on a palatable food for mole crickets. When applied in August, baits were found to be more effective than other modes of application (Short and Koehler 1977). Among common insecticides, fipronil (Chipco Choice, Bayer Environmental Science) is the most effective.

Life Cycle and Biology of Scapteriscus vicinus and S. borellii

Mole crickets eggs are dirty brown or grayish, which are initially white, oval and unsculptured. In the egg chamber, females lay a clutch of eggs, average number of 35 per clutch. Once eggs are laid, females usually do not revisit the chamber, unlike *Neocurtillia hexadactyla* females, which do revisit the chamber. In central Florida, egg laying for both species begins by the latter half of March and is mostly completed by the first of July (Hayslip 1943). Every activity of the mole crickets from flight period to oviposition and egg hatch will show differences due to geographic location, and hence may not be the same in places with different latitudes. Incubation of eggs is temperature dependent. Incubation is shortest when soil temperatures are around 29 °C (84°F) and longest when soil temperatures around 20 °C (69 °F). Incubation is typically 5 d shorter for *S. vicinus* (Hayslip 1943).

Eggs hatch into nymphs, which are similar in appearance to adults, except they lack wings. In central Florida, hatch of *S. vicinus* occurs during late April, while eggs of *S. borellii* hatch in early May (Hayslip 1943). Once emerged, nymphs begin searching for food, feeding on eggs, chorions, and even weaker nymphs. Nymphs molt 6–10 times before becoming adults. The different molts can be recognized by pronotal length which ranges from 1.81-8.14 mm (0.07–0.32 in) (Hudson 1987). Number of molts and number of days required for becoming adults varies between species. The development of wingpads is another character associated with the development stages in mole cricket. The three stages of wingpad development are absence of wingpads, presence of small wingpads, presence of large wingpads. One or two of the penultimate nymphal stages will have small wingpads whereas the last nymph stage has large wingpads (Hudson 1987) . Southern and tawny mole crickets are univoltine. However, a portion of the population may become adults before winter, whereas most overwinter as nymphs (Hayslip 1943).

Cooler soil temperatures cause them to remain underground until favorable conditions allow them to start their nuptial flights. In a mark and recapture study, one mole cricket was collected 8 kms from the release site, although they are not typically long distance fliers (Frank et al. 1994). Adult mole crickets have two pairs of wings which are covered by a network of veins, which are tough tubes supporting the wing membranes. Spaces between the veins are called cells. Forewings of males and females differ slightly in that forewings of males have a pair of large cells. The anterior sides of these cells are harp-shaped and called the stridulatory area. Females lack such large cells. In males, one of the veins on each wing is modified with a line of tiny teeth to form a stridulatory file. This file is drawn across a scraper on the other wing. As the wings close, this makes a sound, which is their mating song (i.e. stridulation). Males repeat this song by opening and closing their wings in rapid succession. The arrangement of the teeth in the stridulatory file differs from species to species, so the song differs from species to species. The loudness of the songs varies but is typically about 70 dB at 15 cm from the source. The song is amplified by widening the mouth of their gallery into a funnel shape. The hearing organs (tympanum) of mole crickets are on the tibiae of the forelegs (Frank et al. 1994).

Male tawny and southern mole crickets sing at night to attract females. Flights begin soon after sunset and end after about one hour in both the tawny mole cricket and southern mole cricket (Hayslip 1943, Ulagaraj 1975). There are two flight periods for both southern and tawny mole crickets; a major spring flight and minor fall flight both of which occur when high temperatures are over 18 °C (Ulagaraj 1975). In fall, however, southern mole crickets are predominantly found (Hayslip 1943). The peak of tawny mole cricket flight is March to early April, and Southern mole cricket flight is April to early May (Hudson 2001). About 14 d after mating, the female lays eggs (Hudson 2001).

Natural Enemies and Biological Controls

In the native range, *Scapteriscus* mole crickets are uncommon and rarely of pest status (Hudson et al. 1988). This is likely due to the presence of natural enemies which keep their populations in check. Similarly, certain predators have been noted for attacking mole crickets in the United States. Subterranean predators like ground beetles from the genus *Pasimachus* and assassin bugs of the genus *Sirthenea* attack adults and nymphs. Adults of bombardier beetle species, *Stenaptinus jessoensis* (Morawitz) are specialist predators of mole cricket eggs (Hudson et al. 1988). Other ground dwelling predatory insects and spiders known to attack mole crickets include earwigs (Dermaptera), ground beetles (Carabidae), tiger beetles (Cicindellidae) and spiders in the families Lycosidae and Salticidae (Hudson et al. 1988). Two *S. vicinus* captured in acoustic traps in New Orleans were found to be parasitized by *Acridomyia* spp. (Diptera: Anthomyiidae) which are usually parasitoids of grasshoppers (Acrididae) (Henne and Johnson 2001). Birds like sandhill cranes and cattle egrets forage on mole crickets when they are near to the surface. In some instances, foraging of insectivorous animals like armadillos do more damage to turf than the mole crickets themselves. Among other vertebrate predators are foxes, skunks, raccoons, toads, and birds (Sailor 1984) which await them above ground.

Apart from these predators, mole cricket populations are subjected to attack by pathogens like bacteria, fungi, virus and protozoans. Fungal pathogens like *Beauveria bassiana* (Balasamo), *Isaria* spp., *Metarhizium anisopliae* (Metchnikoff), *Entomophthora* spp., and *Sorosporella uvella* have been found to infect mole crickets (Hudson et al. 1988). Entomopathogenic nematode *Steinernema* (=*Neoaplectana*) *carpocapsae* Weiser which attacks a wide range of insects were also found to infect mole crickets (Lima Costa et al. 1984).

Despite this list of endemic or naturalized arthropods and pathogens which attack *Scapteriscus* mole crickets, their populations were never significantly reduced. In Florida, a mole cricket task force was formed to investigate classical biological control of mole crickets using natural enemies from their native range. The classical biocontrol agents which have been successfully introduced and established in Florida are the red-eyed fly *Ormia depleta* (Diptera: Tachinidae), beneficial nematode *Steinernema scapterisci,* and the wasp *Larra bicolor* (Sphecidae). In Florida, acoustic trap captures of *Scapteriscus* mole crickets over 25 years on the same sites before and after these biological controls were established indicate a 99% reduction in trap captures (Frank and Walker 2006). This has been attributed largely to the success of the introduction of these biological controls.

In June 1939, *Scapteriscus* mole crickets collected from Belem, Brazil were found to be parasitized by a tachinid fly (Wolcott 1940). In 1983, gravid females of the same species responded to synthesized calls of *Scapteriscus borellii* in Paraguay (Fowler and Kochalka 1985) earlier reported (Mangold 1978). This fly was first identified as *Euphasiopteryx australis* (Wolcott 1940) and later found to be *Euphasiopteryx depleta* (Sabrowsky 1953). The generic name was later made a synonym of *Ormia*, thus changing the name to *Ormia depleta* (Wood 1987). After collecting the fly and hosts from Piracicaba, Brazil, the first laboratory reared colony was achieved in 1987. In spring 1988, adult *Ormia depleta* were released in Florida (Frank et al. 1996). These flies have been successfully reared on all species of *Scapteriscus* mole crickets and on two gryllids (*Anurogryllus* and *Gryllus*) (Fowler and Mesa 1987). Extensive research and releases of the tachinid fly in Florida succeeded in establishing local populations. In just one year, there was an 85% decrease in mole cricket infestations compared to areas where the fly had not become established (Frank et al. 1996). By the summer of 1994, *O. depleta* had spread throughout Florida and this spread was associated with decreases in mole cricket populations (Frank et al. 1996). Acoustic traps in New Orleans, LA have trapped *S. borellii* which were parasitized at a greater rate than was earlier reported from Florida (Frank et al. 1996, Henne and Johnson 2001).

In 1985, *Steinernema scapterisci* was brought from Uruguay to Florida. After laboratory evaluation, the first small plot releases were done near the University of Florida in Alachua County in June 1985 (Parkman et al. 1993). There are three separate life stages: egg, four stages of juveniles, and adult (male and female) of which the third stage juvenile is the most important for biological control of mole crickets. They enter the mole cricket through its mouth or spiracles. Once inside, the juveniles release bacteria into the hemocel of their host. The bacterium causes death of the mole cricket by multiplying within, and the nematode feeds on the bacteria (Nguyen and Smart 1992). Initially released as a classical biocontrol agent or biopesticide, this nematode has been commercialized and now sold under the trade name Nematac S™. The successful inoculative release of this exotic entomopathogenic nematode was the first classical biocontrol against mole crickets (Parkman et al 1994, Parkman and Smart 1996). Even though the initial costs of using Nematac S is considerably higher than the cost of an insecticide treatment, infection may persist from year to year. Even though studies have shown that mole crickets can behaviorally avoid soils treated with entomopathogenic fungi (Villani et al. 2002), after *Steinernema* were established, up to 80% of mole crickets trapped on those sites were infected (Leppla et al. 2007). This nematode is also compatible with insecticides commonly used in turfgrass (Barbara and Buss 2006),

***Larra bicolor*- Mole Cricket Killers**

Over 60 species of the genus *Larra* (Hymenoptera: Sphecidae) are known to be parasitoids that prey on mole crickets (Bohart and Menke 1976). *Larra americana* Saussure now known as *Larra bicolor* (Bohart and Menke 1976) was introduced into Florida through the efforts of the mole cricket task force. Successful introduction (Wolcott 1938) and establishment (Wolcott 1941) of *Larra bicolor* into Puerto Rico triggered the search for this natural enemy to control the damage caused by pest mole crickets.

Morphology of Larra bicolor

The abdomen of *Larra bicolor* is solid red and wings are smoky brown or indigo blue with silvery white markings on their head. In contrast, *Larra analis*, a native species that attacks *Neocurtilla hexadactyla,* has similar colored wings and a dark colored abdomen with only red at the tip (Figure 1.4).

The tarsal claws are short and without inner teeth. The foretibia has spine rows, without teeth on inner margin of mandible and propodeum is constricted at the level of spiracles with a glossy integument. Females have a thicker pronotal collar, straight on top and as high as scutum and sternum II without a pair of special flattened oval areas basally. The pygidial plate is bare and without a transverse row of short setae apically. Females are generally bigger (average size 22 mm) than their male counterparts (Frank and Sourakov 2002).

Figure 1.4

a. *Larra bicolor* b. *Larra analis* (photo credit E. Trammel, AR)

Parasitism and Biology of Larra bicolor

In shorter mown turfgrass, female *Larra* wasps make short flights across the grass landing then initiating a walking search for hosts (personal observations). Most host searching behaviors observed in Mississippi were on cloudless days with low to moderate winds (i.e, ≤ 10 mile per hour), which is consistent with Wolcott (1938). Also, *Larra* are generally first observed in Mississippi in early to mid morning continuing through the late afternoon if conditions are favorable (personal observations). There is variation reported in the foraging behavior of *Larra* wasps observed by different researchers in different locations. In Brazil, flight abundance was relative to the beginning and end of the rainy season (February and May, respectively). This is attributed to the ease of locating host tunnels in the moist soil during these times. Soil and ambient air temperatures may also play a role in hunting behavior. As soil temperatures increased to 50°C, host hunting became sporadic (Castner and Fowler 1987). When wasps are not hunting they may seek shelter in the foliage of nectar producing plants or behavior may switch from hunting to nectar feeding.

When hunting, *Larra* wasps walk on the surface of soil antennating the soil and turf as they move (personal observations). When a surface tunnel is encountered the wasp will enter and exit the same tunnel many times after which, due to unknown reasons, the mole cricket rushes out of the tunnel, followed by the wasp (Castner and Fowler 1987). Although attacks may occur underground, the wasp is more frequently observed attacking prey on the soil surface. It is reported that nearly half of prey that exit the hole escape parasitism (Castner and Fowler 1987). The wasp stings the mole cricket and lays an egg once the prey is in hand. This wasp is an ectoparasitoid, and its venom only temporarily paralyses the mole cricket providing a window for oviposition. After laying the egg, the wasp may leave the mole cricket (Frank and Sourakov 2002) or remain to guard it until the host recuperates from paralysis and resumes its normal routine (Castner 1988). In studies, it was observed that majority of the wasps laid only one egg on the ventral side of the mole cricket. If an egg was earlier laid, it would be removed before another was laid. This behavior enhanced the chance of the single larva successfully developing (Castner 1986).

Egg incubation may take as little as 4 days in summer, and there are 5 larval instars before pupation. Time from oviposition to pupation is temperature dependent and takes from 12–30 days (Cushman 1935, Frank and Sourakov 2002). About 2 weeks after oviposition, the wasp larvae now 14–26 mm, will kill the host, consuming the soft body parts leaving only the sclerotized portions. The cocoon is formed in about a day and is constructed of a silken case and commonly covered with sand. The adult wasp emerges 6–8 weeks after cocoons are formed (Cushman 1935, Frank and Sourakov 2002).

Host Specificity of <u>Larra</u> <u>bicolor</u>

Larra bicolor will successfully develop on all species of *Scapteriscus* mole crickets present in the United States but not on the native species, *N. hexadactyla*. The native mole cricket when attacked emits a sticky anal secretion which entangles the wasp, not allowing further parasitism. Even when *L. bicolor* can readily oviposit on *N. hexadactyla*, the wasp larva cannot successfully develop on that host. *Larra analis*, the aforementioned native species, is the common parasitoid of *N. hexadactyla* (Frank and Sourakov 2002). Having coexisted since early 1900s, the native species, *Larra analis* have not yet been considered a parasitoid of *Scapteriscus* mole crickets.

Nectar Sources for Larra bicolor

Larra bicolor is reported to feed on flowers of several species of unrelated weeds and wildflowers including dollar weed and ragweed (Frank et al. 1994). A review of literature shows there have been no studies to identify ornamental plants with flowers that can be used by *L. bicolor*. Among the weedy species, *Spermacoce verticillata* (Rubiaceae) is preferred by the wasp and is commonly used to monitor wasp activity (Meagher and Frank 1998). Floral characteristics important for *L. bicolor* are floral depth, which is equal to the length of their glossa in case of *Spermacoce* (Arevalo and Frank 2005), and a nectar with a sucrose: hexose ratio >1 (Arevalo and Frank, unpublished data).

Populations of *L. bicolor* on flowers are male biased in the morning, when presumably females are hunting, but contain more females in the afternoon and early evening (Castner and Fowler 1987). Flower feeding or foraging occurs from 0830 to 1530 hours for male and female *L. bicolor* (Castner and Fowler 1987) but is dependent on temperature. In addition to feeding, these wasps may also rest on flowering plants. The origins or purpose of such behaviors are not known.

CHAPTER II

SEASONAL AND DIEL ACTIVITY OF *LARRA BICOLOR* IN THE SOUTHEASTERN UNITED STATES

Abstract

Even though *Larra bicolor* has been controlling mole crickets in Florida since 1993, there has been no study to document the seasonal and diel activity of the wasp in North America. The only previous study (Castner and Fowler 1987) that documented the diel activity was conducted in Puerto Rico or its native range and hence may be different from wasp behaviors in North America. The objectives of this study were to record the seasonal activity of the wasp in the northern Gulf Coast region and to record the diel activity of foraging wasps. Areas in south Mississippi where populations of *Larra bicolor* were known to be established were surveyed every two weeks during 2006 - 2008 to document the seasonal activity. In one of these areas, hourly observations were done to note the diel activity of the wasp. Wasps were typically active from late May to early December with slight variations between locations. Wasps were active as early as 0600 h and remained active until 1900 h. This information is useful to incorporate an integrated approach to control mole crickets by reducing insecticides and timing sprays in such a way that wasps are not affected.

Introduction

Most of the pre-release research, including biological and ecological studies on *Larra* wasps, was conducted in Brazil and Puerto Rico. The most widely cited study on the seasonal flight period and diel patterns of *L. bicolor* was conducted in Puerto Rico. This study, conducted in one geographic location over 10 d in a single growing season, does not provide information about the activity of this wasp in subtropical parts of North America. As the wasp expands further north and west in the southern U.S. (Held 2005, Abraham et al. 2008), it is important to understand its flight activity in this region. The objective of this study hence was to monitor the seasonal activity and diel activity of *Larra bicolor* in the Northern Gulf region.

Materials and Methods

From June–August 2006, eight golf courses in south Mississippi were visited and surveyed for *L. bicolor*. Many of these courses include sites in south Mississippi where *Larra* was originally discovered (Held 2005). Of these eight, three (Table 2.1) were selected for seasonal monitoring of *Larra bicolor*. On these three courses, two sites where *L. bicolor* were found on more than one occasion were selected for monthly monitoring.

Table 2.1

Locations Selected for Monthly Monitoring of *Larra* Wasps

Name, Location	**Site 1**	**Site 2**
Gulf Hills Golf Club, Ocean Springs, MS	**5th fairway**	**7th Tee**
St. Andrews Golf Club, Ocean Springs, MS	**5th fairway**	**7th green**
Great Southern Golf Club, Gulfport, MS	**11th fairway**	**Back 9**
MS State University, Coastal REC, Biloxi, MS	**Flower garden**	**Flower garden**

From April–December, 2006–2008, each site was scouted for 15–20 minutes for wasp activity. On each golf course site, areas infested with mole crickets and showing fresh activity were first scouted. These areas were selected because *L. bicolor* commonly walks over the turf searching for active burrows in which to enter. Observations done in the garden at the Coastal REC were of wasps foraging on flowering plants (i.e., *Spermacoce verticillata* or *Pentas lanceolata*). Since these wasps are noted nectar feeders, monitoring flowering plants would gauge the activity of *L. bicolor* in that location. Monitoring was done on clear days, between 1000–1500 hours. Observations were done every two weeks.

Diel activity of *L. bicolor* was studied by monitoring wasp activity on flowers of blooming plots of *Spermacoce verticillata* and *Pentas lanceolata*. These plots were part of the garden experiment described in more detail in Chapter III. This study was conducted from 7–10 Aug, 2007 and 9-11 Oct, 2008 when wasp populations were abundant enough to collect data. The number and sex of wasps on flowering plants in eight plots in the garden were taken hourly from 0500–2000 h each day. Female and male wasps were distinguished based on morphological differences of their wings. The female *Larra* wasp had a solid black color to its wing and a dark red abdomen opposed to the light red abdomen and wing lighter at the base and darker at the apex in the males (Figure 2.1)(personal observations).

Figure 2.1

a. *Larra bicolor* Female b. *Larra bicolor* Male

During these experiments, observations were made for 20 minutes each hour. Wasps that landed on flowers were counted even if they departed without actually feeding. Data were collected all day despite changes in weather such as rain or overcast conditions. Temperature data was taken from data loggers (#450, Spectrum Technologies, Plainfield, IL) adjacent to the flower garden.

Results

The seasonal activity of *Larra bicolor* has been monitored since 2006 and is in its third year. Typically, wasps were first observed in May and activity continued until December (Figure 2.2) when the ambient temperatures typically were at or below 20°C. Wasps were not observed on golf courses browsing on the turf or parasitizing mole crickets before June or after November. In the flower garden, however, wasps were present earlier and later in the season. Wasps were active on all sites surveyed except one. Wasps were present at St. Andrews golf course in 2006 but never observed thereafter.

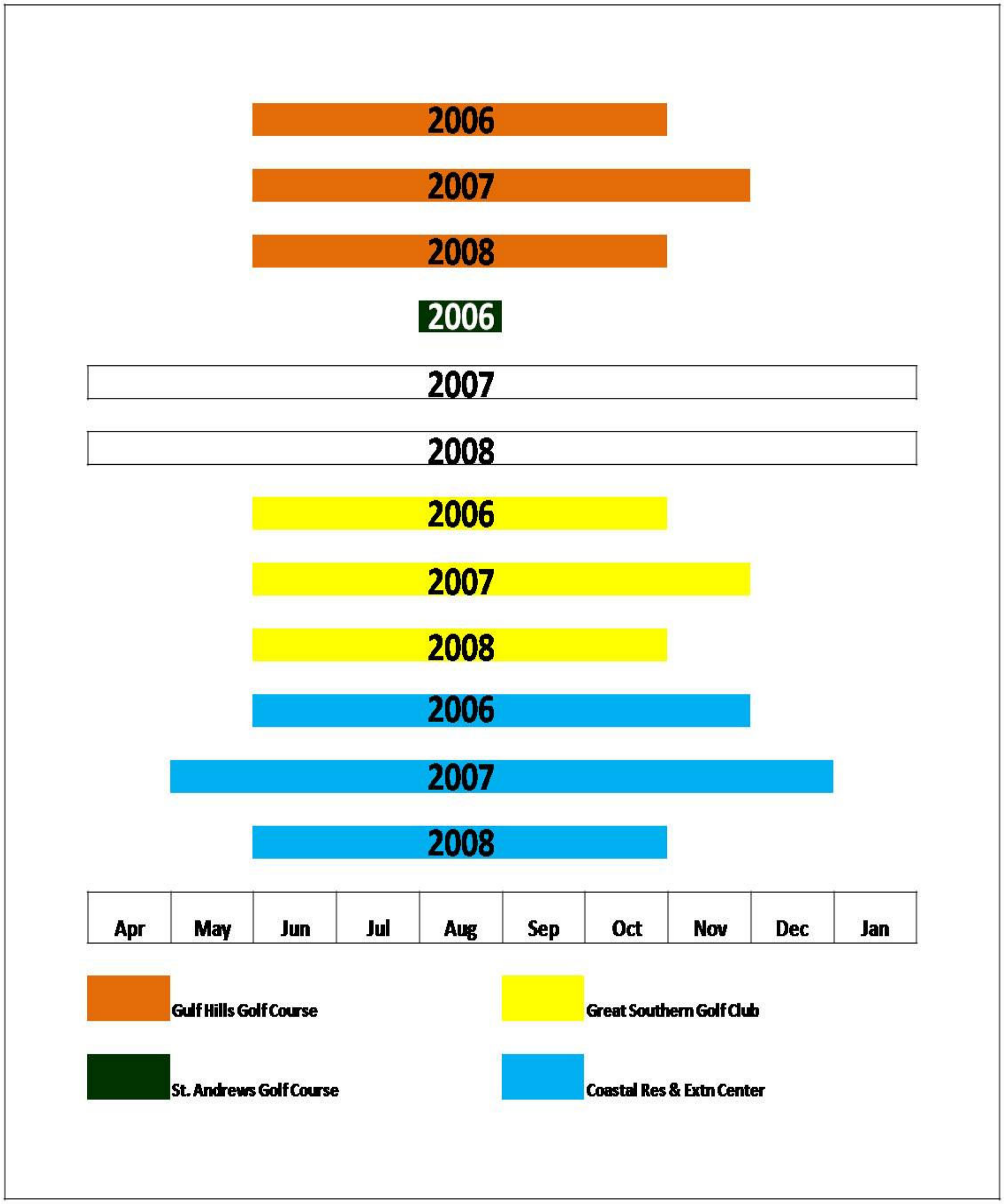

Figure 2.2

Seasonal Activity of *Larra bicolor*

Wasps were observed on flowers from about 0600 h to 1900 h each day in summer and from 0900 h to 1700 h in fall when temperatures were cooler (Table 2.2). Sunrise and sunset during summer days were at 0520 hrs and 1845 hrs, and 0600 hrs and 1730 hrs during fall, respectively. Males were consistently present at each hourly observation whereas female foraging was more sporadic. Peak foraging on flowers is hard to determine because some wasps could remain on flowers and not forage. Wasps were still counted if present on the flowers from the previous observation. The greatest number of males was recorded (Table 2.2) at 1800, 1000, and 1000–1100 hours for Aug 7–9, respectively, where as the greatest number of males during Oct 9-11 were at 1500 hrs on all three days. The greatest number of females was recorded at 1100, 1300, and 1000 for Aug. 7–9, respectively and during Oct 9-11 abundance of females was also around the 1500 hr mark. Wasps were observed resting in the shaded interior of flowering plants and just a few were observed to spend the overnight period on the plant. Interestingly, most wasps leave the plant at or near dusk. Attempts to follow individual wasps at the end of the day were not successful. Wasps could be “Sleeping” in surrounding vegetation in a cluster or alone as seen in other solitary wasps (O' Neill 2001).

Table 2.2

Diel Activity of *Larra bicolor*

Date	Time	Number of wasps				
		Male	Female	Total	Soil Temperature (°C)	Air Temperature (°C)
08-07-07	0500	0	0	0	28.7	27.6
	0600	0	0	0	28.7	27.2
	0700	1	6	7	28.3	27.2
	0800	1	0	1	28.3	28.3
	0900	7	0	7	28.3	30.4
	1000	14	0	14	28.3	31.6
	1100	15	0	15	28.7	34.1
	1200	11	3	14	28.7	34.9
	1300	16	**14**	30	29.5	34.9
	1400	12	6	18	29.9	31.2
	1500	1	4	5	28.7	24.4
	1600	5	9	14	28.0	26.0
	1700	25	5	30	28.0	29.5
	1800	**31**	9	**40**	28.3	31.2
	1900	15	5	20	28.3	30.4
	2000	0	0	0	28.3	29.5
08-08-07	0500	0	0	0	28.0	27.6
	0600	0	0	0	27.6	26.8
	0700	14	6	20	27.6	27. 2
	0800	7	0	7	27.6	28.3
	0900	13	2	15	27.6	29.9
	1000	**21**	7	**28**	28.0	31.6
	1100	7	1	8	28.0	34.1
	1200	17	1	18	28.7	35.8
	1300	18	**10**	**28**	29.5	33.6
	1400	9	7	16	29.5	34.1
	1500	8	2	10	29.9	34.9
	1600	3	0	3	30.4	33.6
	1700	2	0	2	30.4	33.6
	1800	2	0	2	30.4	32.8
	1900	4	0	4	29.9	32.4
	2000	0	0	0	29.9	32.0
08-09-07	0500	0	0	0	28.7	28.0
	0600	0	0	0	28.3	27.6
	0700	0	0	0	28.3	27.6

Table 2.2 Contd…

	0800	0	0	0	28.3	29.5
	0900	9	0	9	28.3	31.6
	1000	**18**	**11**	**29**	28.7	32.8
	1100	**18**	5	23	29.1	34.9
	1200	4	1	5	29.5	34.9
	1300	6	1	7	29.9	34.5
	1400	3	5	8	30.4	34.9
	1500	1	0	1	30.4	29.1
	1600	4	0	4	30.4	31.6
	1700	0	0	0	29.9	29.1
	1800	1	0	1	29.5	27.6
	1900	0	0	0	29.5	28.3
	2000	0	0	0	29.1	28.0
10-09-08	0500	0	0	0	23.7	16.0
	0600	0	0	0	23.7	16.0
	0700	0	0	0	23.3	16.4
	0800	0	0	0	23.3	16.8
	0900	1	0	1	23.3	18.3
	1000	1	1	2	23.3	20.6
	1100	0	2	2	23.7	21.3
	1200	2	0	2	24.4	21.7
	1300	10	1	11	24.8	24.0
	1400	**15**	0	15	25.6	24.8
	1500	**15**	**3**	**18**	26.4	25.2
	1600	6	1	7	26.8	24.0
	1700	1	1	2	26.8	23.3
	1800	0	0	0	26.8	23.7
	1900	0	0	0	26.4	20.2
	2000	0	0	0	26.0	19.0
10-10-08	0500	0	0	0	23.7	16.8
	0600	0	0	0	23.3	16.4
	0700	0	0	0	23.3	16.4
	0800	0	0	0	23.3	17.1
	0900	0	0	0	22.9	19.4
	1000	1	0	1	23.3	22.5
	1100	0	1	1	23.7	25.2
	1200	5	0	5	24.4	26.8
	1300	19	**2**	21	25.2	27.2
	1400	16	**2**	18	26.0	28.3
	1500	**23**	**2**	**25**	26.8	28.7
	1600	13	0	13	27.2	28.3

Table 2.2 Contd…

	1700	5	1	6	27.2	27.2
	1800	0	0	0	27.2	25.2
	1900	0	0	0	27.2	22.1
	2000	0	0	0	26.8	21.3
10-11-08	0500	0	0	0	24.0	18.3
	0600	0	0	0	24.0	18.3
	0700	0	0	0	24.0	18.3
	0800	0	0	0	23.7	19.0
	0900	0	0	0	23.7	22.5
	1000	1	0	1	24.0	24.8
	1100	2	0	2	24.4	26.8
	1200	13	0	13	25.2	26.4
	1300	19	**2**	21	26.0	28.3
	1400	23	1	24	26.4	27.6
	1500	**25**	**2**	**27**	26.8	28.0
	1600	14	0	14	27.2	27.2
	1700	2	0	2	27.2	25.6
	1800	0	0	0	27.2	24.8
	1900	0	0	0	26.8	23.3
	2000	0	0	0	26.4	23.3

* Numbers in bold denote the peak activity during the day

Discussion

This study is the first to document daily foraging activity of *L. bicolor* in North America. Previous studies on the diel behavior were conducted in Puerto Rico and Brazil. Foraging activity of *L. bicolor* wasps in Puerto Rico was between 0830 and 1530 hours (Castner and Fowler 1987) notably shorter than the daily foraging times reported here in summer. Maximum activity occurs with soil temperature between 28-30°C and an air temperature of 30-35°C. Even though wasp population at nectar sources seem to be male biased, generally around 1300–1400 hrs, male: female ratios on flowers are nearly one to one. Males and females forage on flowers to obtain nectar (carbohydrates) to sustain activity. Females have a more ephemeral presence due to actively searching for hosts for oviposition. Castner and Fowler (1987) also reported male biased populations on flowers. Males were never observed to attempt to mate while females foraged which is consistent with observations by Menke (1992) that mating occurs on the grass. Wasps were observed resting in the shaded interior of flowering plants and just a few were observed to spend the overnight period on the plant. Resting in the shade may indicate a thermoregulation behavior to cool the thoracic temperature because such behavior was noticed from 1200 hrs to 1400 hrs when the temperature was at its peak during the day. Wasp left the plants between 1800–1900 h in summer and 1700 - 1800h during fall, but it is not known where these wasps spend the night period. Because air temperatures did not seem to trigger this, wasps may use dusk (1845 h) as a trigger to leave the nectar sources.

Because there is no passive monitoring system for the wasp, direct observations are the only means of monitoring. This in part explains why data from an entire month were pooled and not presented as separate observations. In June 2007, for example, there was a 2 week period when no wasps were found. This has been called the ‘summer slump’ (H. Frank, personal communication) and describes a gap between the emergences of two generations. St. Andrews Golf Course reportedly had wasps in 2004 (Held 2005) and some were noted in 2006. However, all subsequent surveys failed to show the wasps. The only reported change during this time is a more strict insecticide regime which is evident in less mole cricket activity on the course. The impact of turfgrass insecticides on *Larra bicolor* has not been studied.

CHAPTER III

ORNAMENTAL PLANTS AS NECTAR SOURCES FOR *LARRA BICOLOR*

Abstract

Parasitic wasps generally benefit from access to carbohydrates, primarily in nectar from flowering plants. *Larra bicolor* feed on nectar of certain wildflowers and weeds including *Spermacoce verticillata*. The objective of this study was to evaluate certain ornamental plants as nectar sources for adult *L. bicolor* and to evaluate the potential benefits of flower feeding on longevity and parasitism. Plants similar to *Spermacoce* in structure and nectar source were used to find if any of them attracted *Larra bicolor*. A common garden experiment including 13 taxa of ornamental plants and *S. verticillata* in four replicates was used to determine the preference of *L. bicolor* for certain flowering plants under field conditions. *Pentas* attracted as many wasps as *Spermacoce*. A white-flowered variety of pentas was visited more often by *L. bicolor* than other pentas varieties. These results were further investigated in laboratory longevity tests. Wasp survival on Pentas was comparable or better than survival of wasps provisioned with *Spermacoce*. These results suggest that pentas may be planted in landscapes to sustain this wasp and perhaps enhance biological control.

Introduction

Flower feeding is common in parasitic hymenopterans (Jervis et al. 1993) directly consuming floral and extrafloral nectar or pollen, to a lesser extent trichomes, epidermis or indirectly honeydew. Presence of nectar or other carbohydrate sources can attract and retain natural enemies, (Evans and Swallow 1993, Jacob and Evans 1998) thereby increasing parasitism. Incorporating flowering plants adjacent to turfgrass has implications for pest management. *Tiphia vernalis* (Hymenopetera: Tiphiidae), introduced for biological control of Japanese beetle (*Popillia japonica*) grubs, were found to prefer flowers of *Peonia* spp. in gardens with other blooming plants (Potter and Rogers 2004). In landscapes or on golf courses, decreased pest and increased predator populations were obtained when conservation strips and wildflower mixes were incorporated (Braman et al. 2002, Frank and Shrewsbury 2004). Similarly, parasitism of Japanese beetles grubs by *Tiphia vernalis* was greater within 1 m of a patch of flowering *Peonia* spp. (Potter and Rogers 2004).

Similar patterns are visible in *Larra bicolor*, an ectoparasite of exotic *Scapteriscus* mole crickets, which notably visits flowering plants such as *Spermacoce verticillata* (Rubiaceae). The presence of nectar sources are reportedly important to establishment of *L. bicolor* (Leppla et al. 2007). Aravelo and Frank (2005) evaluated four species of flowering wildflowers, *Conoclinium coelestinum*, *Elephantopus elatus*, *Passiflora coccinea*, and *Solidago fistulosa* for visitations by *L. bicolor* and sugar content of their nectar. All were visited less than *S. verticillata* and had a lower sucrose-fructose ratio.

Flowering ornamental plant species commonly grown for aesthetics or for attracting butterflies are already present in urban landscapes. Identifying flowering plants that would also provide an ecological service may facilitate establishment and success of *L. bicolor* in lawns and golf course turfgrass. Since *S. verticillata* is a weed in coastal areas (Everest et al. 1996) (Figure 3.2) and overwinters in the landscape in coastal Mississippi (personal observations), the objective of this study was to investigate alternative ornamental plants with flowers of similar shape and size as *Spermacoce*, particularly other species in the Rubiaceae.

In south Mississippi, *L. bicolor* was found feeding on *Pentas lanceolata* (Rubiaceae) in the landscape (personal observations Figure 3.1). This suggests that pentas and perhaps other flowering plants in the Madder family could substitute as a nectar source for *L. bicolor*. Varieties of *P. lanceolata* vary in flower color and plant height. Pentas are common in southern landscapes for their heat tolerance and attractiveness to butterflies (Bruner et al. 2002). Because pentas are similar in flower shape and in the same family as *Spermacoce*, other members of the Madder family may also be attractive to *L. bicolor*.

Figure 3.1

Larra bicolor Feeding on *Pentas lanceolata*

Despite the previous research on *L. bicolor*, the influence of nectar sources on wasp survival or parasitism are not known. Leppla et al. (2007) speculates that "The wasps (*L. bicolor*) will forage for mole crickets more effectively if *S. verticillata* plants are available as a source of nectar". This hypothesis and the preference of *L. bicolor* wasps for ornamental plants in the field were tested to determine the influence of nectar sources on wasp longevity in the laboratory.

Figure 3.2

Spermacoce verticillata as a Weed on the Roadsides of Florida

Materials and Methods

Source of Plants and Wasps

Plants for this study were grown at the South Mississippi Branch Experiment Station in Poplarville, MS or purchased from local retailers. The plants were not treated with insecticides but were fertilized using 10-50-10 (Schultz Co, Bridgeton, MO) regularly to keep them blooming. Wasps for use in these experiments were field collected within 12 h of the start of the experiment. Wasps were collected using a battery-powered vacuum modified for insect collection (# 2820A, Bioquip Products, Rancho Dominguez, CA) or collected using an aerial net. Wasps were stored in a cooler with ice packs for transport to the lab.

Evaluation of Flowering Plants in a Common Garden Experiment.

In May 2007 and in 2008, a garden of potential nectar sources was established on the grounds of the Coastal Research and Extension Center, Biloxi, MS. Each plant species or cultivar (Table 3.1) was established in four replicated plots, 0.5 × 0.5 m. This entire garden was covered with about 5 cm of ground hardwood mulch to reduce weed populations and drip irrigation lines established throughout.

Beginning in late May each year, plants were inspected for *L. bicolor*. Data were collected when more than 2-3 wasps were in the garden. Numbers of wasps on flowering plants were recorded between 1100–1200 h. Fifteen minute observations were made in each replicate. Wasps landing on the flowers were counted even if they did not actively feed. Plants species visited and the number of foraging wasps was recorded and summarized over 6 d in 2007 and 10 d in 2008. Numbers were then analyzed using

Poison Analysis to compare the relative number of visits to certain ornamental flowers and determine the preference of *Larra bicolor*. Plots where plants were not flowering were excluded before the analysis.

Table 3.1

Horticultural Plants Evaluated as Nectar Sources for *Larra bicolor*

Achillea millefolium	*Pentas lanceolata* 'Butterfly Pink'	Fennel
Agastache urticifolium	*Pentas lanceolata* 'Butterfly White'	*Verbena* 'Aztec'
Anethum graveolens	*Pentas lanceolata* 'Butterfly Red'	*Verbena bonariensis*
Cephalanthus occidentlis	*Asclepias* 'Silky Gold'	*Verbena* × *hybrida* 'Blue Lagoon'
Ixora coccinea	*Asclepias* 'Silky Red'	*Verbena* × *hybrida* 'Lady Scarlett'
Lobularia maritime	*Bouvardia* spp	*Verbena* × *hybrida* 'Quartz Waterfall'

.

Figure 3.3

The Garden Experiment with Plants in Bloom

Evaluation of Wasp Longevity in the Laboratory.

Laboratory experiments were conducted on 11–27 August, 2007 to determine the longevity of adult *L. bicolor* provisioned with flowers. These experiments were conducted in 22.7 liter, plastic cylindrical test arenas (Sterilite, Townsend, MA, Figure 3.4). These arenas were 43.81 cm tall to accommodate a small potted plant. The bottom of each area was filled with dry sand to a depth of 8 cm. An empty plastic pot was placed in the center so that plants could be easily added. A single field-collected wasp was introduced to each arena. In arenas where flowering plants were absent a sugar diet or no diet was provided. The sugar diet was a 0.1% sugar solution which was dabbed on a cotton ball and placed on a plastic platter. This was changed every other day to make sure that sugar water was available. Wasp survival was evaluated daily. The mean number of days wasps survived in each treatment was analyzed using analysis of variance (Statistix, Analytical Software, Tallahassee, FL) followed by Tukey's means separation procedure ($P < 0.05$).

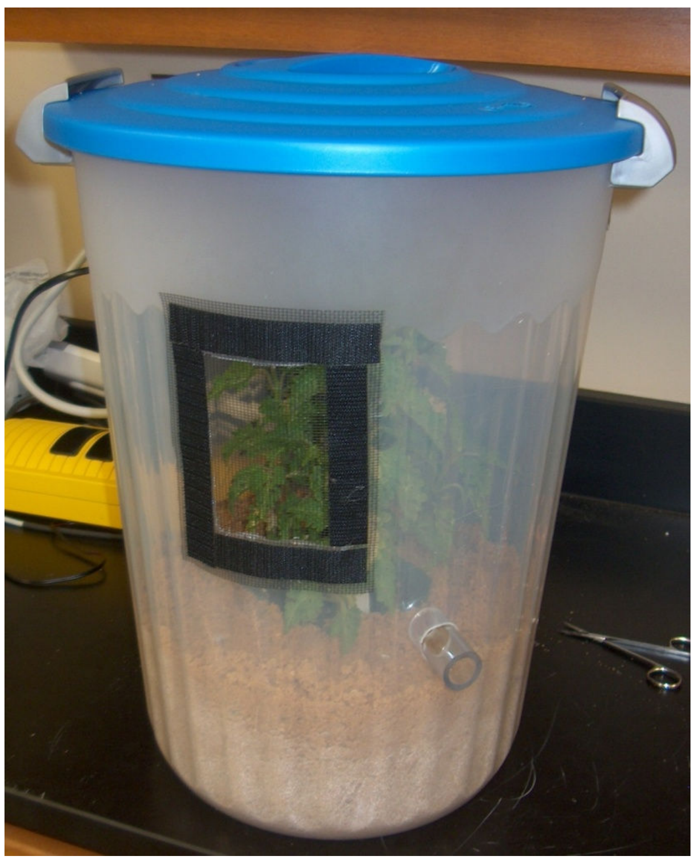

Figure 3.4

Arena for the Longevity Tests

Results

Evaluation of Flowering Plants in a Common Garden Experiment.

In both years, *L. bicolor* wasps were only observed on flowers of *Pentas* and *Spermacoce.* All varieties of pentas regardless of flower color were visited by *L. bicolor.* Among these, significantly more wasps visited white-flowered pentas and Spermacoce (Figure 3.5, F = 12.09, $P < 0.0001$). Ability of recruiting wasps was similar for both White Pentas and *Spermacoce.*

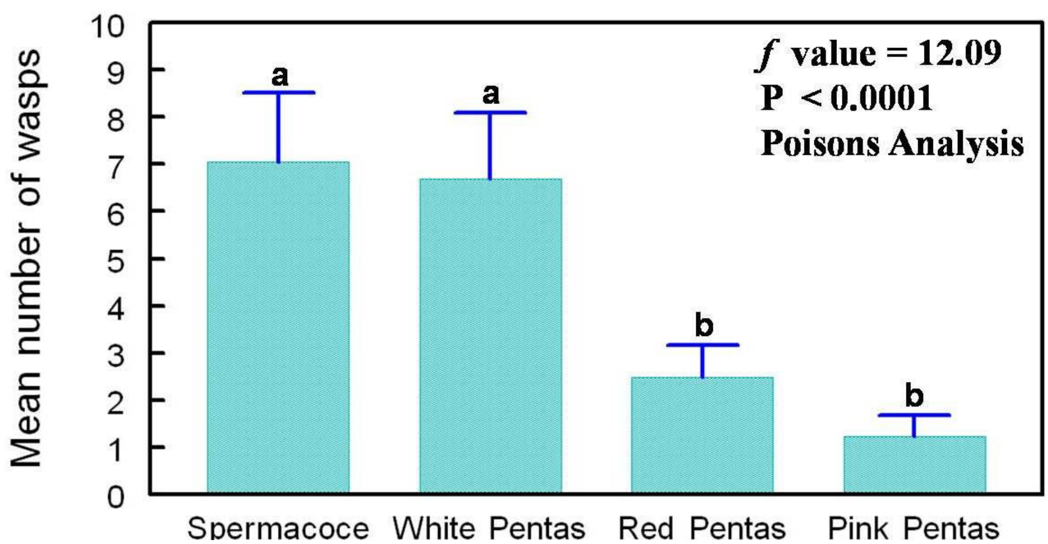

Figure 3.5

Field Test Results of Ornamental Plant Preference for *Larra bicolor*

Laboratory Longevity Assays.

Trial I: Wasps held in captivity with no food survived, on average, 1.75 d which was the same as those provisioned with flowering Spermacoce. Wasps provisioned with flowering White Pentas lived 5d longer than those provisioned with Spermacoce (F = 10.12, $P = 0.012$). Wasp longevity ranged from 0–8 days in this experiment.

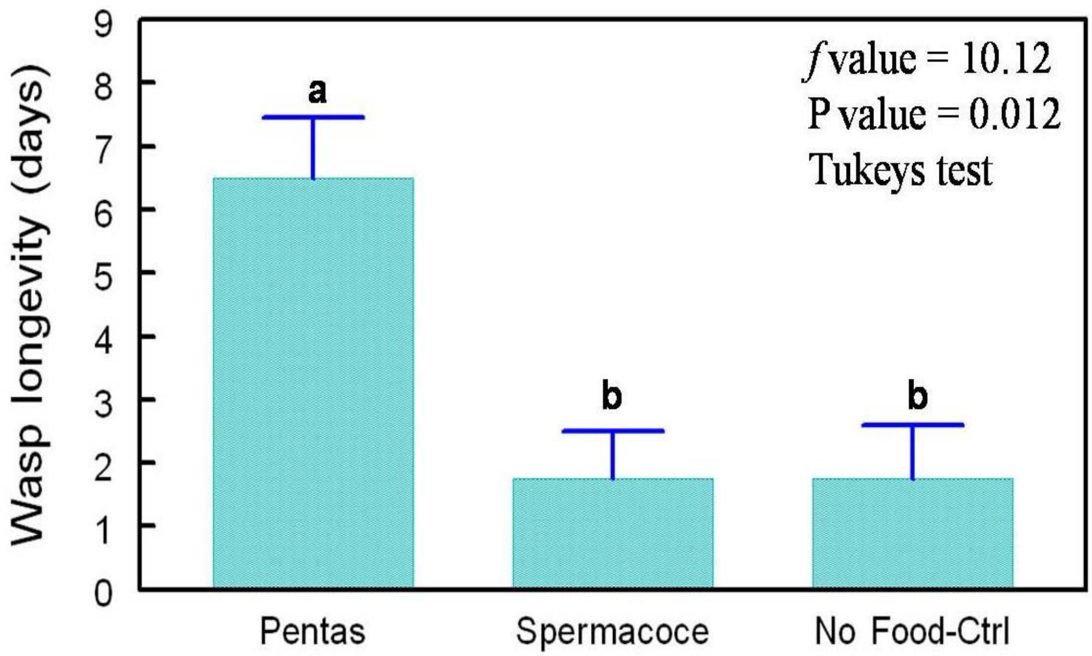

Figure 3.6

Lab Trial I - Choice Test with No Food Control, *Spermacoce* and *Pentas*

Trial II: There were no significant differences between treatments in this experiment indicating that all three food sources are comparable (F = 1.1, $P = 0.39$).

Wasp longevity ranged from 2–6 days in this experiment. The average longevity of males and females were 4.2 and 4.3d, respectively.

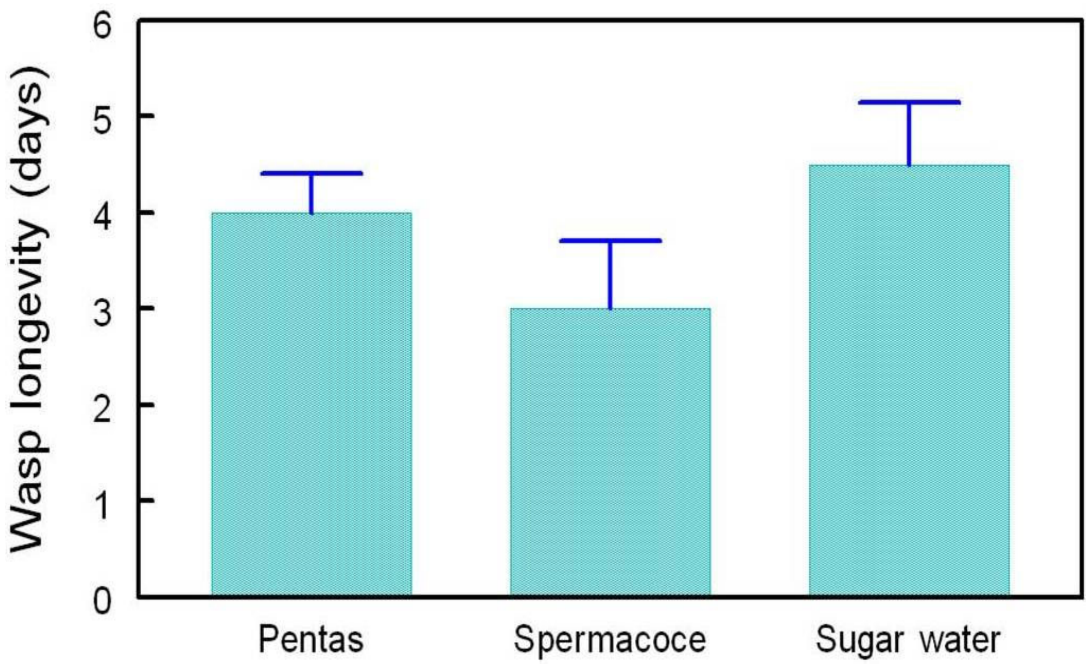

Figure 3.7

Lab Trial II with *Spermacoce*, *Pentas* and Sugar Water Control

Discussion

Flower morphology, color, or sugar content of the nectar may explain the similar response of wasps to pentas and Spermacoce. Spermacoce is the primary nectar source for *L. bicolor* wasps in Florida. This species is listed as a weed (Everest et al. 1996) and numerous volunteers of *S. verticillata* in areas adjacent to the garden plot have required chemical control. Fortunately, field choice tests suggest that an ornamental species, Pentas, may be a suitable replacement. The glossal length of male and female *L. bicolor* are not different and roughly equal to the flower depth of *S. verticillata* (Arevalo and Frank 2005). In the field experiment, there was no difference in numbers of wasps recorded on pentas with white flowers and Spermacoce. Flower morphology (i.e., size) likely has limited importance since individual pentas flowers are larger than flowers of Spermacoce. Color alone does not explain preference as wasps have been observed feeding on plants with a range of flower colors (Arevalo and Frank 2005).

Results of laboratory trials provide additional evidence that *Pentas* is comparable or perhaps better than *Spermacoce* relative to wasp longevity. Nectar is an excellent source of carbohydrates (sugars) for insects. Similar field preference research with *Pentas* and butterflies (Bruner et al. 2002) also suggests that *Pentas* has nectar volume or composition which makes it attractive to nectar feeding insects. Wasp flowers and butterfly flowers tend to be sucrose rich (Baker and Baker 1983). *Spermacoce* has sucrose-dominated nectar (H. Frank, unpublished data). Deep-tubed flowers tend to have sucrose rich nectar (Baker and Baker 1983) and may explain the similarity for *Pentas* and *Spermacoce* in recruiting wasps even though *Spermacoce* has small flowers compared to *Pentas*.

Even though plants in the same family could show similarities in nectar content, that need not be the case in large families (Baker and Baker 1983) and this explains why other plants in the madder family did not recruit the wasp. This is indication that floral characteristics like floral volatiles or nectar composition would be the key to wasp preference. Since color variants of the same plant tend to have similar nectar content (Baker and Baker 1983) the ability to recruit wasps by the other colors of *Pentas* could be explained as a function of the nectar content. The difference in wasp attraction due to color need not be due to difference in nectar content.

CHAPTER IV

FIRST REPORT OF *LARRA BICOLOR* (HYMENOPTERA: SPHECIDAE) IN ALABAMA

Abstract

Larra bicolor is a parasitic wasp introduced into Florida for biological control of pest mole crickets. Wasps hunt for mole crickets in the pastures and managed turfgrass. Once the wasp finds a mole cricket, they chase it to the surface and stings it. The sting temporarily paralyzes the mole cricket enabling the wasp to deposit an egg. The wasp egg hatches and larva feeds as an external parasite. The larva consumes the mole cricket then spins a cocoon in the soil. There are 2-3 generations of *L. bicolor* each year. Wasps have spread throughout Florida and into the northern Gulf states such as Mississippi where *L. bicolor* were discovered in 2004. The present study was conducted to determine if *L. bicolor* was present on golf courses in Mobile and Baldwin Counties (southernmost counties) in Alabama. In fall 2007, eight golf courses in these counties were identified and surveyed for *L. bicolor*. Wasps were observed or collected on 4 of the 8 golf courses surveyed. The discovery of *L. bicolor* in southern and coastal Alabama provides a new record for this species but more importantly it provides new information for turfgrass managers. On-going research projects are investigating the role of flowering plants in

parasitism and survival of *L. bicolor*. Turfgrass managers and homeowners in Alabama can now utilize the findings of this and future research on biological control of mole crickets.

Introduction

Mole crickets (Orthoptera: Gryllotalpidae: *Scapteriscus* spp.) are serious pests of warm season turfgrass and pastures in the southern United States. Since their entry into United States along the Georgia coastline in the early 1900's, *Scapteriscus* mole crickets have spread south into Florida and west through Louisiana. In Mississippi and the northern Gulf region, two species, *S. vicinus* and *S. borellei*, are present whereas these two and *S. abbreviatus* are present in Florida. Damage from *Scapteriscus* mole crickets is most severe in the southern most areas of Gulf states (Frank and Walker 2006). On golf courses, insecticides are used extensively to prevent damage from mole crickets. For example, control may cost up to $675 per ha. Combined effects of three natural enemies, *Ormia depleta* (Diptera: Tachinidae), *Steinernema scapterisci* (entomopathogenic nematode), and *Larra bicolor* F. (Hymenoptera: Sphecidae) are attributed with a 99% reduction in mole cricket populations in Florida (Frank and Walker 2006).

Figure 4.1.

a. *Larra bicolor* (15–20 mm long) is Shown Here Feeding on Flowers of *Spermacoce verticillata* b. Stinging a Mole Cricket.

Larra bicolor (Figure. 4.1) is an ectoparasitoid of mole crickets and has 2 to 3 generations per season (Frank and Sourakov 2002) . The biology and ecology of *L. bicolor* were recently reviewed (Frank et al. 1995) and will not be discussed in detail. *Larra bicolor* wasps are black with wings that are brown to blue and a solid red abdomen (Figure. 4.1) which distinguishes it from *Larra analis* F. which is native to Mississippi and other Gulf states, and has a black abdomen with red just at the tip (Menke 1992). Between 1981–1983, researchers collected *L. bicolor* wasps from Puerto Rico and released them at five sites in Florida. Following these releases, wasps only became established at the southernmost release site near Ft. Lauderdale (Frank et al. 1995). In an attempt to establish populations in northern Florida, three species of wasps, *Larra bicolor*, *L. praedatrix*, and *L. godmani* were collected from Bolivia and released along with parasitized hosts at three sites near Gainesville in 1988-89 (Frank et al. 1995).

Again, these releases appeared unsuccessful until 1993 when *L. bicolor* were observed attacking mole crickets and feeding on flowers of *Spermacoce verticillata* in Gainesville.

In 2004, several specimens of *L. bicolor* were collected from golf courses in coastal Mississippi. *Larra bicolor* currently is widespread in Florida (Frank et al. 1995, Frank and Walker 2006) and reported from southern counties in Mississippi and Georgia (Held 2005). The most northern collection of *L. bicolor* in Mississippi is (30.69°N) Saucier, MS (D.W. Held, unpublished data) and most established populations of *L. bicolor* in the U.S. occur at ≤ 31°N latitude (Frank and Walker 2006). *Larra bicolor* was intentionally released in southern Georgia in 2000 by W. Hudson for biological control of mole crickets (W. Hudson, personal communication). With no records of release in Mississippi, the discovery of *L. bicolor* suggests natural range expansion of this wasp along with its host (Held 2005). Range expansion from Florida to Mississippi would indicate that *L. bicolor* may also be present in Alabama. For this reason, we sampled golf courses in the southern and coastal areas of Alabama to determine if and where *Larra bicolor* was present. If found, turf managers in south Alabama would benefit from knowing *L. bicolor* is present and could utilize on-going research to enhance local populations and parasitism.

Materials and Methods

For this survey, southern Alabama (< 31°N latitude) was divided into three regions; the metro Mobile area, the eastern shore of Mobile Bay, and coastal Alabama including Gulf Shores. Between 26 Sept and 19 Oct. 2007, golf courses in each region were sampled for *L. bicolor*. On each course, the greens staff or superintendents suggested at least three fairways infested with mole crickets to sample. Surveys were conducted between 0800–1400 hours. All observers walked, side by side, in a line spanning the width of the fairway. Every observer walked each fairway from tee to green. Particular attention was given to areas with fresh mole cricket damage. If *L. bicolor* wasps were observed attempts were made to collect them. When captured, wasps were collected with nets then transferred to collection bottles in a cooler for transport to the laboratory. If not captured, the wasp was noted as observed but not collected and the survey resumed to the end of the fairway.

Results and Discussion

Larra bicolor wasps were found on at least one golf course in each of the three geographical regions in south Alabama (Table 4.1). This record is the first report of this beneficial wasp in Alabama. It is probable that the populations of *L. bicolor* established in south Alabama have resulted from natural range expansion of this wasp within its host's range (Held 2005).

Adult *L. bicolor* was rarely found in all areas surveyed on a particular golf course. Wasps were more common in areas or on golf courses where fresh mole cricket damage was more apparent. Some golf courses were more aggressive with their mole cricket management programs and those courses had fewer or smaller areas that were damaged. Because *L. bicolor* wasps are difficult to sample, non-detection of wasps on certain sites during this survey should not be equated with an absence of wasps on that course or geographical area. In a survey of golf courses in coastal Mississippi in 2004, Held (2005) reported certain sites where *L. bicolor* were present or absent. Since that survey, *L. bicolor* has been collected from one site where it was undetected in 2004, and populations are now absent from one site where they were once present (Held and Abraham, unpublished data). Temporal or seasonal abundance in wasp populations is likely determined by host abundance, presence of nectar sources (Leppla et al. 2007), or surface-applied insecticides.

Table 4.1

Numbers of *Larra bicolor* Reported from Golf courses in South Alabama.

Golf course	**Observed**	**Collected**
Metro Mobile (Mobile Co.)		
Spring Hill College	9	3
Country Club of Mobile	0	0
Azalea City	3	1
Eastern side of Mobile Bay (Baldwin Co.)		
Timber Creek	0	0
Quail Creek	12	4
Coastal (Gulf Shores and Orange Beach)		
Kiva Dunes	0	0
Peninsula	8	0
Golf Club at the Wharf	0	0

Figure 4.2

a. A Female *L. bicolor* Preparing to Enter the Mound in Search for a Mole Cricket. b. Mole Cricket Mound with Hole that was Commonly Observed on Sites where *L. bicolor* Wasps were Active.

On three of the four sites where *L. bicolor* wasps were found, mole cricket mounds with holes in the top of them were also observed (Figure. 4.2 a, b). *Larra bicolor* females move along the grass surface searching for mounds. Once a mound is detected, the soil is antennated and the wasp may choose to enter. To enter a mound, the female must burrow through the soil at the surface. It then proceeds into the subterranean tunnels in search of a host. As a result, mole crickets commonly exit the mound to avoid attack. Holes similar to these are commonly observed on several sites in south Mississippi where *L. bicolor* are present. These holes, therefore, may result from either the wasp entering or the mole cricket escaping the mound and may be a passive indication that *L. bicolor* is active in that area.

The discovery of *L. bicolor* in southern and coastal Alabama provides a new record for this species but more importantly it provides new information for turf managers. On-going research projects in Florida (H. Frank, personal communication) and Alabama are investigating the role of flowering plants in parasitism and survival of *L. bicolor*. Turf managers in Alabama can now utilize the findings of this and future research on biological control of mole crickets.

CHAPTER V

CONCLUSIONS

Detection of *Larra bicolor* in Mississippi in 2004 indicated that the wasp expanded its range from Florida without any human interference. Our subsequent detection of *L. bicolor* in Alabama supports this concept of natural range expansion. In some Alabama sites, we observed greater numbers of wasps than what are seen in one location in Mississippi, which suggest populations may have first established in Alabama.

One of the reasons attributed to the success of the wasp establishment in Florida has been the availability of its nectar source *Spermacoce verticillata* which is a weed in southern turf. Field and lab studies we conducted indicate that *Pentas* could sustain the wasp for longer times than *Spermocace*. This provides another reason to include *Pentas* in our landscape at least where mole crickets are a problem.

Seasonal activity in the northern gulf region is enough for 2-3 generations, and will be acting on a single generation of mole crickets that emerge in a year. The diel activity though was more interesting, when it showed two distinct patterns, when they sporadically appeared on the nectar sources during summer and when they had a peak activity period in the fall when the temperatures reached the maximum in the day before their activity again ceased around sunset. These observations indicate that the activity of the wasps is dependent on a combination of sunrise, sunset timings and temperatures.

With the information this study provides, we are better equipped to utilize the services of *Larra bicolor* as a biocontrol agent as we know that that they are available in southern parts of Mississippi and Alabama. We know that *Pentas* can substitute *Spermacoce* as a nectar source and with knowledge about its daily and seasonal activity, we could time our insecticide applications later in the day, and even reduce part of it to lower the costs of control and also to augment wasp populations.

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